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Title:

Optomagnetic detection of microRNA based on duplex-specific nuclease assisted target recycling and core-satellite magnetic superstructures

Authors & affiliations:

Bo Tian,[†] Zhen Qiu,[†] Jing Ma,[‡] Teresa Zardán Gómez de la Torre,[†] Marco Donolato,[§] Mikkel Fougt Hansen,[#] Peter Svedlindh,[†] Mattias Strömberg[†]

[†] Department of Engineering Sciences, Uppsala University

[‡] Department of Immunology, Genetics and Pathology, Uppsala University

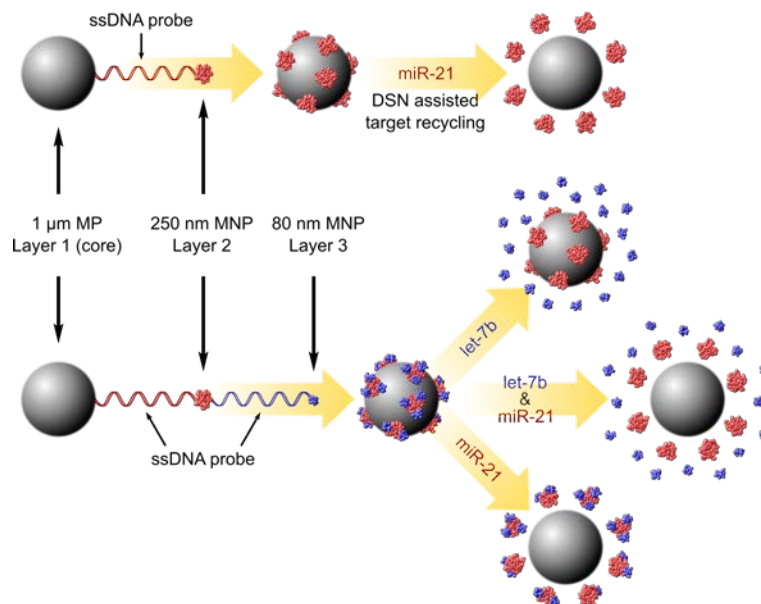
[§] BluSense Diagnostics

[#] Department of Micro- and Nanotechnology, Technical University of Denmark

Superstructural assembly of magnetic nanoparticles (MNPs) enables biosensing by combining specially tailored properties of superstructures and the particular advantages associated with a magnetic or optomagnetic read-out such as low background signal, easy manipulation, cost-efficiency and potential for bioresponsive multiplexing.

Herein, we demonstrate a miRNA detection method based on optomagnetic readout, duplex-specific nuclease (DSN) assisted target recycling and the use of a core-satellite magnetic superstructure consisting of streptavidin MNPs linked to biotinylated target-binding ssDNA strands linked to microparticles. Triggered by the presence of target miRNA and DSN-assisted target recycling, the core-satellite magnetic superstructures released MNP satellites to the suspension where they could subsequently be accurately quantified in the optomagnetic setup. Target miRNAs were preserved in the cleaving reaction and could thereby trigger further release of MNPs. The optomagnetic method detected spectra of the 2nd harmonic modulation of the intensity of 405 nm laser light transmitted through the MNP suspension in response to an applied oscillating magnetic field. The target-triggered release of MNPs was detected as a turn-on of the Brownian relaxation response from free MNPs.

For singleplex detection of let-7b, a linear detection range between 10 fM and 10 nM was observed and a detection limit of 4.8 fM was obtained within a total assay time of 70 min. Multiplexing was achieved by releasing nanoparticles of different sizes in the presence of different miRNAs. The proposed method also has the advantages of single-nucleotide mismatch discrimination and that it works in a clinical sample matrix.



We further utilized on-particle rolling circle amplification (RCA) to improve the preparation of core-satellite magnetic superstructures. The on-particle RCA based core-satellite magnetic

superstructures had a higher load of satellite MNPs and faster response compared to ssDNA probe based core-satellite magnetic superstructures. By using on-particle RCA, the detection sensitivity was improved to 1 fM.

